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**The effect of sodium and carbohydrate in a rehydration food on
subsequent exercise performance**

by

Huimin Yan

A thesis submitted to the graduate faculty
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Major: Kinesiology (Biological Basis of Physical Activity)

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Ames, Iowa

2008

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ABSTRACT

The purpose of this study was to determine the effectiveness of two rehydration beverages on fluid restoration and metabolism on subsequent exercise following exercise- and heat-induced dehydration. Twelve male subjects were dehydrated by ~3 % of body mass with exercise and heat exposure. In a randomly assigned, counterbalanced design, subjects were rehydrated at room temperature with either 175 ml of chicken noodle soup (SOUP: Campbell Soup Company, Camden, NJ) or an artificially sweetened placebo (CON) at the beginning of the 120 min rehydration period and 20 min later. Water was ingested every 20 min during the remaining 100 min of rehydration. Soup contained 161.0 mmol/l Na⁺, 5.3 mmol/l K⁺, and 32.6 g total carbohydrate. CON contained 14.4 mmol/l Na⁺, 16.0 mmol/l K⁺, and no carbohydrate. Total fluid ingestion was matched with body fluid loss during dehydration. After rehydration, subjects performed 30 min of steady state exercise at 68% VO₂peak. Subjects then performed a time trial in which they accumulated as rapidly as possible the amount of work equal to 30 min of exercise at 70% VO₂peak in a thermo-neutral environment (25 C and 40% relative humidity).

There was no significant difference in percent recovery of body mass during rehydration (CON: 79.4 ± 3.3%; SOUP: 80.9 ± 3.4%). Time trial performance was significantly improved in SOUP (30.6 ± 0.8 min) compared with CON (33.2 ± 1.4 min; p=0.031). The rate of carbohydrate oxidation tended to be higher in SOUP (2.33 ± 0.09 g/min) compared with CON (2.18 ± 0.11 g/min; p=0.076). No differences in heart rate or ratings of perceived exercise were found during post-rehydration exercise. These results

suggest that ingesting chicken noodle soup after exercise in the heat improves subsequent endurance exercise capacity, possibly through enhanced CHO oxidation.

CHAPTER 1. INTRODUCTION

Overview

During exercise in the heat, heavy sweating can cause a substantial loss of body water and electrolytes. Although extracellular fluid and intracellular fluid both contribute similar absolute volumes of water of fluid loss during dehydration, extracellular fluid experiences greater relative loss, resulting in a decreased plasma volume (8).

If fluid loss was not fully restored, dehydration has been shown to impair cardiovascular function and thermoregulation during subsequent exercise (12). Sodium is thought to be important in restoration of volume loss after dehydration. Sufficient electrolytes need to be ingested to maintain plasma osmolality (33). Ingesting large amount of water with low concentrations of electrolytes may increase urine loss due to fluid-regulating hormones such as aldosterone, rennin and angiotensin II (21, 32). Carbohydrate and sodium accelerate water absorption in the small intestine membrane through co-transporters (21).

Although a wealth of studies have examined rehydration after heat and exercise induced dehydration, most studies have not assessed the effectiveness of rehydration beverages on subsequent exercise performance. The ingestion of carbohydrate during rehydration period has also shown to improve subsequent exercise (16, 47). Exercise time to exhaustion was improved after CHO intakes ranging from 50 g to 220 g and large amount of water.

Most hydration studies have examined either water or carbohydrate-electrolyte beverage. Only a few studies have employed food during rehydration period. Previous research found a standard meal and water provides quicker rehydration than a sports drink

(29). A previous study in our lab investigated the effect of different rehydration fluids or meal (34). In each trial, 350 ml of water, chicken broth, a carbohydrate-electrolyte drink, and chicken noodle soup was given at the early state of rehydration and water was given during the rest of 2 h rehydration period. By the end of rehydration period, plasma volume remained significantly below predehydration values in water and CE trials. In contrast, plasma volume was not statistically different from predehydration values in chicken broth and chicken noodle soup trials. However, exercise capacities after rehydration were not examined in these previous studies. It is possible that the improvement in plasma volume restoration observed in soup trials would promote post-rehydration exercise capacities. Another subject of interest is whether soup would change substrate utilization during rehydration and post-rehydration exercise.

Statement of the Problem

The purpose of this study was to investigate the effectiveness of two different rehydration beverages: water (CON) and chicken noodle soup (SOUP) on improvement in exercise performance after rehydration.

CHAPTER 2. REVIEW OF LITERATURE

Water and Electrolytes

Water (H₂O) is the most abundant component of the human body, constituting ~60% of body mass. Body H₂O is distributed between two major fluid compartments: the fluid within the cells, intracellular fluid (ICF), and the fluid surrounding the cells, extracellular fluid (ECF). Extracellular fluid can be divided into plasma, interstitial fluid, lymph and transcellular fluid. The volume and percentage of body fluid are provided in Table 1.

Table 1. Classification of body fluid

Compartment	Volume of Fluid (in Liters)	Percentage of Body Fluid (%)	Percentage of Body Mass (%)
Total Body Fluid	42	100	60
Intracellular Fluid (ICF)	28	67	40
Extracellular Fluid (ECF)	14	33	20
Plasma	3	6.6 (20% of ECF)	4
Interstitial fluid	11	26.4 (80% of ECF)	6
Lymph	Negligible	Negligible	Negligible
Transcellular fluid	Negligible	Negligible	Negligible

Fluid Shift and Electrolyte Loss during Exercise

Extracellular fluid and ICF both contribute to the fluid loss during exercise. In one study, plasma volume (PV) decreased 12% in the first 10 min of endurance exercise. Despite heavy sweating for an additional 110 min of exercise, PV only dropped additional 3.6% (9, 10). In a related study, Costill et al. (9) observed an increase (8%) in water content in active

muscles during the first minutes of exercise, with an equivalent decrease in PV. The water content in the inactive muscle was slightly decreased at the same time. Bergstrom et al. (2) also found that heavy exercise caused water accumulation in active muscles, accompanied by lactate accumulation. It was suggested that the accumulation of water in active muscles was related to the increased osmolality caused by the production of lactate. Therefore, the flux of water to active muscles from plasma occurred at the beginning of exercise may be driven by osmotic gradient. The slight additional decrease in PV with additional 110 min of exercise (10) may also be explained by the changes in osmolality. In the same study, plasma osmolality increased throughout the 2 h exercise bout. Water content in active muscle gradually decreased along with the slight decrease in PV during the 110 min of exercise while inactive muscle water remained constant (8). It was suggested that plasma hyperosmolality may induce a return of water to the intravascular space. These studies suggest that the fluid shift in the body during exercise was largely driven by the plasma osmolality (2, 8-10).

Costill et al. (8) measured fluid loss compartments at different exercise and heat-induced dehydration levels. At lower levels of dehydration (2% body mass loss), roughly 70% came from ECF and 30% from ICF. Plasma contributed to approximately 10% of total body water loss at three dehydration levels. At 4% and 6% dehydration, intracellular water contributed to 52% and 50% of total water loss, respectively. Although ICF and ECF contributed similar absolute volumes of water, the ECF component experienced relatively greater loss of water.

Bergstrom et al. (2) observed that intracellular potassium concentration decreased after heavy exercise. However, the decrease in potassium was accompanied by an

accumulation of muscle water and no appreciable shift of potassium across the cell membrane. Therefore, the decreased potassium concentration was mainly due to dilution. Intracellular sodium concentration increased. There was evidence that hydrogen ions were actively transported out of the cells (36). The increase in sodium concentration was suggested to be linked with an electrolyte pump common to sodium and hydrogen ions (36).

Na⁺ and fluid restoration

Heat and/or exercise induced dehydration may adversely affect exercise performance (12). To effectively restore body fluid becomes a challenge to people who need to recover from dehydration. The addition of Na⁺ in rehydration beverages has been intensively studied (12, 27, 28, 32, 33). Costill et al. (12) showed the ingestion of a carbohydrate-electrolyte drink produced lower urine output than plain water, and higher plasma restoration was observed in carbohydrate-electrolyte drink trials. In contrast, Ray et al. (34) found no difference in plasma volume restorations after rehydration with CHO-electrolyte drinks (Gatorade) and water intake. However, in the CHO-electrolyte drink trial, only 350 ml of CHO-electrolyte drink was ingested at the beginning of rehydration, and the rest of beverage was made up by water. In previous studies, the rehydration beverages were only CHO-electrolyte drinks. Different rehydration protocols with previous studies (12) may explain the discrepancies in rehydration effectiveness.

The concurrence of higher osmolality and better rehydration observed in rehydration with water and sodium capsules implies osmolality is critical in rehydration process (33). It has been proposed that plain water alone might increase urine loss of sodium and water; plasma rennin activity and aldosterone levels might be responsible for these effects (32).

These studies along with other studies (27, 30) showed rehydration after exercise-heat induced dehydration is effective when both water and sufficient electrolytes were ingested (28).

A number of studies have assessed the effect of different sodium doses in rehydration beverages. Mitchell et al. (30) found that the participants achieved only 73% rehydration after 3 h of rehydration with 14 mmol/L Na^+ despite the total fluid is 150% of fluid lost during dehydration. Maughan and Leiper (27) dehydrated participants by 1.9% body mass by intermittent cycle exercise in a warm, humid environment. The participants were rehydrated with beverages of different sodium contents in a volume of 150% of fluid loss during the following 5.5 h. In a rehydration drink with 26 mmol/L sodium content, the participants only retained 53% of fluid intake, suggesting a negative fluid balance of 79.5%. In another drink with 2 mmol/L sodium, participants were still in negative fluid balance (54%) at the end of 5.5 h. However, 52 mmol/L and 100 mmol/L Na^+ produced positive fluid balance of 103.5% and 110% respectively. A meta-analysis suggested that to achieve full recovery of fluid deficit within 6 h, rehydration beverages should contain 100 mmol/L sodium if total rehydration fluid volume equals the fluid lost during dehydration. If the rehydration fluid is 150% of fluid loss, 50 mmol/L sodium will be required to balance fluid deficit (39).

A group of researchers tested the interaction between Na^+ content and fluid volume (41). Participants in this study were dehydrated by approximately 2% of body mass. Rehydrating with 23 mmol/L Na^+ drink, even though the total fluid intake was 150% or 200% of body mass loss, body fluid were not completely balanced after 6 h. In contrast, when rehydrated with a higher sodium content (61 mmol/L), total fluid intake equal to 150% of body mass loss produced positive fluid balance after 6 h. These studies suggest when sodium

levels are relatively low, even large quantities of fluid do not appear adequate to produce rapid rehydration.

Therefore, ingestion of merely water is not enough to restore exercise induced dehydration (41). If a large quantity of plain water was ingested after exercise and heat induced dehydration, plasma sodium concentration and plasma osmolality would drop. And rehydration will not be effective because large volume of fluid ingested will be lost through urinary output.

Na⁺ and fluid-regulation hormones

Fluid-regulating hormones such as antidiuretic hormone (ADH, also known as AVP/arginine vasopressin) play important roles in urine production and thus rehydration effectiveness. Antidiuretic hormone is released upon stimulus of hypothalamus osmoreceptors. When ECF osmolarity increases, ADH is released by hypothalamus to increase reabsorption in distal and collecting tubules in the kidney. Aldosterone secretion is increased due to low ECF volume and low arterial blood pressure. Aldosterone is the most important factor in regulation ECF volume because of the aldosterone-controlled Na⁺ reabsorption mechanism. When ECF osmolality decreases, the release of ADH is depressed.

Extracellular Na⁺ plays an important role in osmolality determination. It has been discussed previously that rehydration with water or diluted electrolytes solution will result in lower plasma osmolality (32, 33). A reduction in the concentrations of fluid regulatory hormones such as aldosterone, angiotensin II and renin (21, 32) accompanied by low plasma osmolality might explain higher urine volumes and less restoration of body fluid in water or diluted electrolytes solution groups.

Carbohydrate may also play a role in influencing rehydration after exercise or heat-induced dehydration. Data from several studies demonstrated that the addition of small amount of glucose to water had an inhibitory effect on gastric emptying (11, 24). On the other hand, fluid replacement depends on both gastric emptying and intestinal absorption. Adding 10% CHO into rehydration solutions was shown to be effective in restoring fluid deficit (12). The mechanisms associated with the effective fluid restoration may be explained by improved intestinal water absorption, since the absorption of water through the small intestine membrane is accelerated by the active co-transport of sodium and CHO (21). Therefore, to ensure effective rehydration, the concentration of CHO should be balanced to minimize the adverse effect on gastric emptying and maximize the desired improvement on intestinal water absorption.

Plasma glucose and muscle glycogen

Endurance for exercise at intensities between 70-80% VO_2max can be increased by elevated muscle glycogen stores and decreased by lowered muscle glycogen. In a study, muscle glycogen levels were found depleted after exercise at 1000 kpm/min for 20 min (2,45). One of the subjects in the study was not able to finish 20 min of exercise and terminated exercise at 17 min of exercise. Muscle glycogen level of this subject decreased from 25.4 to 2.6 mmol/ 100 g fat free solid. These studies lead scientists to believe that amount of glycogen present is an important determinant of the capacity for continuous muscular work. And back in the 1960s, scientists also believed that muscle glycogen was the primary carbohydrate source during exercise.

The normal blood glucose is approximately 5.5 mmol/L. During prolonged endurance exercise, maintenance of blood glucose homeostasis becomes a challenge, because skeletal muscle changes from little glucose uptake to a situation requiring greatly increased glucose demand. A study observed glucose uptake by the leg increased 7-fold above resting levels during light exercise (400 kg.m/min) and rose 10- and 20-fold at exercise intensities of 800 and 1200 kg.m/min, respectively (45).

A study examined substrate usage during prolonged strenuous exercise (74% $\text{VO}_{2\text{max}}$) with and without CHO feedings (13). Fatigue developed 3 h later when fed with a placebo. Although fatigue was delayed by 1 h when fed CHO (2.0 g/kg BM at the beginning and 0.4 g/kg every 20 min thereafter), the pattern of decline in muscle glycogen concentration was similar in the first three hours. During the first two hours of exercise, CHO oxidation rates were similar in both trials. A decline in CHO oxidation was observed during the third h of the placebo trial when plasma glucose concentration decline significantly to 2.5 mmol/l at the point of fatigue. In contrast with the previous acknowledgement about the importance of muscle glycogen, although muscle glycogen were low at the end of the 3 h exercise in both trials, subjects in the CHO feeding trials were able to maintain plasma glucose at 4-5 mmol/l and exercise 1 additional hour. It was suggested by the authors that blood glucose can largely replace muscle glycogen in providing CHO for oxidation in latter stage of prolonged strenuous exercise when muscle glycogen level are low. Hypoglycemia causes muscular fatigue when muscle glycogen is low. It was suggested that the ingested CHO probably replaced liver glycogen as a source of blood glucose during the first 2 h of exercise.

There is a concurrence of increased hepatic glucose output and maintenance of circulating blood glucose (45, 48). The mechanism to stimulate hepatic glucose output has

been suggested to be a fall in blood glucose at initiation of exercise (7, 26). Hepatic glucose production increases to match up with glucose needs during exercise. During moderate intensity exercise, hepatic glucose output is regulated by alterations in glucagons, insulin and epinephrine. During moderate intensity exercise, both decrements in insulin and increments in glucagon prevented exercise-induced hypoglycemia by signaling glucose production in the liver (22). Epinephrine is also a factor influencing hepatic glucose output. Epinephrine may be involved in the prevention of hypoglycemia during exercise, at least when the changes in insulin and glucagon do not occur (22). In contrast, during high-intensity exercise, catecholamines, in particular epinephrine, plays an important role in mediating hepatic glucose output (23).

The utilization of muscle glycogen is elevated in a hot environment (20). In one study, six men performed three 15-min exercise bouts at 70 to 85% VO_2max in hot (44 °C, RH = 15%) and cold (9 °C, RH = 55%) environments. Muscle glycogen use by the vastus lateralis muscle was 76% greater in the heat (74 vs. 42 mmol/kg-wet muscle). It has been suggested that the increased muscle glycogen use in the heat is due to the elevated epinephrine level (17). Several studies showed close relationship between intramuscular glycogen use and plasma epinephrine levels during submaximal exercise (18, 19). A more recent study examined muscle glycogen use during submaximal exercise with exogenous epinephrine infusions (17). Epinephrine level was infused to levels similar to the values observed during exercise in the heat. This study indicates that intramuscular glycogen use, glycolysis and carbohydrate oxidation are augmented by elevated epinephrine. This study provides evidence for the causal relationship between elevated epinephrine and increased muscle glycogen use in the heat.

Adding CHO to a rehydration beverage

A number of studies compared the effect of carbohydrate ingestion on exercise performance. In all of these studies, the plasma glucose levels were elevated after CHO ingestion. Some studies reported higher plasma glucose levels after CHO ingestion compared to control (water) trials (4, 38, 47), while others observed higher plasma glucose than baseline, but not different than control groups. In these studies (16, 42), elevated insulin was observed after CHO ingestion.

It should be noted that an elevated plasma glucose level does not necessarily indicate higher CHO utilization. In one study (16), participants performed 90 min of exercises to exhaustion at 70% VO_2max , and they either ingested a 6.9% CHO-Electrolyte beverage providing 1.0 g CHO per kg body mass (CHO group) or placebo solution with equal quantity (Control group) in the next 4 h. After 4 h, exercise to volitional fatigue was performed. During exercise, plasma glucose concentrations were similar in both groups. However, indirect calorimetry based on respiratory data indicated higher CHO utilization in CHO group. Results also showed an average 22.2 min longer in time to exhaustion after rehydration with carbohydrate-electrolyte beverages. Another study supported these results using a similar protocol (47). As noted earlier, blood glucose level during prolonged exercise is mainly maintained by liver glucose output. Wahren et al. (45) found that increased hepatic output of glucose, primarily by means of augmented glycogenolysis, contributed to blood glucose homeostasis in exercise and provided an important source of substrate for exercising muscles. Higher CHO oxidation during exercise may be achieved by higher glucose utilization in the working muscles and higher hepatic glucose output without disturbing blood glucose level.

Two studies did not find differences in the restoration of exercise capacity between CHO intakes ranging from 50 g to 220 g. After a 4 h recovery, ingesting 50 g of CHO resulted in similar run-to-exhaustion time compared to ingesting 169 g of CHO (46). Bilzon et al. (4) found similar exercise time to exhaustion in the group ingesting 55 g of CHO and 220 g of CHO, even though glycogen synthesis was estimated to be approximately fivefold greater in 55 g group. These authors suggested that exercise capacity was limited by thermoregulatory incapacity rather than substrate availability per se. Therefore, adding carbohydrate into rehydration beverage can improve exercise performance in a subsequent exercise bout.

Other factors influencing rehydration

Fluid restoration after dehydration may be influenced by factors other than Na^+ concentration in the fluid (27, 30, 41), volume of fluid ingested (39, 41), CHO concentration (12, 21), and urine production, which are discussed in the previous chapters. There are other factors such as temperature of the fluid ingested (35) and fluid osmolality (11, 44) also affecting the effectiveness of rehydration.

Cold beverages (5°C) empties from the stomach faster than warm (35°C) beverages (11). A later study (35) confirmed the observation that cold fluid produces faster gastric emptying than warm fluid. The time of discharge of an indigestible capsule from the stomach into the duodenum is faster when administered with 250 ml of water at 5 °C than at 20-25 °C or 45 °C. Therefore, cold beverages would be favored to effectively restore fluid loss.

The rate of gastric emptying has been shown to be slowed by an increase in fluid osmolality (11, 44). Costill et al. found that a hypertonic glucose ($\geq 278\text{mmol/L}$) solution

resulted in a marked decrease in gastric emptying (11). Intestinal water absorption has also been found to be smaller using higher osmolality of the test fluid. Shi et al. (40) examined intestinal absorption of three test solutions with different osmolalities: 186, 283 and 403 mOsm/L. Hypotonic test solutions resulted in a 17% higher net fluid absorption than hypertonic solution. However, the advantage of higher net fluid absorption observed in hypotonic test solution did not produce higher plasma volume over the test time. It has to be noted that the subjects in this study were examined under euhydration and plasma volume changes might be different if the study was performed on dehydrated subjects.

Rehydration with food and water

Maughan et al. (29) used either a commercially –available sports drink or a standard meal plus water to rehydrate subjects. Total water intake was 150% of body mass loss during dehydration in both trials. At the end of the 4 h rehydration period, subjects rehydrated with sports drinks were in negative fluid balance, whereas the on the food plus water trials, subjects were euhydrated.

Ray et al. (34) investigated the effect of different rehydration fluids or meal. In a randomized cross-over design, 30 subjects rehydrated with water, chicken broth, a carbohydrate-electrolyte drink, and chicken noodle soup. In each trial, 175 ml of fluid was given at the beginning of rehydration and another 175 ml after 20 min. Water was given during the rest of 2 h rehydration period. Total water consumption was matched with total fluid loss. By the end of rehydration period, plasma volume remained significantly below predehydration values in water and CE trials. In contrast, plasma volume was not statistically different from predehydration values in chicken broth and chicken noodle soup trials.

Exercise and heat induced moderate dehydration can be restored effectively by sufficient ingestion of water and a meal with significant amount of electrolytes. The effect of the meal was as effective as, or even better than a sports drink.

Electrolytes are important in restoring body fluid. However, the purpose of recovery from dehydration is not simply restoring body fluid to euhydrated status. It is also important to replenish endogenous glycogen stores for the subsequent exercise during within short recovery period.

CHAPTER 3. METHODS AND PROCEDURES

Subjects

Twelve healthy male volunteers were recruited to participate in the study. All volunteers signed the informed consent form and were informed of all risks and stress associated with these experiments. They were asked to complete a medical history form. The subjects indicated their past medical history and current health status including physical activity (type and amount) in the medical history form. The subjects were physically active, and they were actively participated in cycling or weightlifting competitions. Peak oxygen uptake ($\text{VO}_{2\text{peak}}$) was obtained while cycling on an electromagnetically-braked cycle ergometer during graded exercise test to the point of exhaustion. Physical characteristics of the subjects are presented in Table 2.

Table 2. Characteristics of the subjects (n=12)

	Age (y)	Height (cm)	Body Mass (kg)	BMI (kg/m^2)	$\text{VO}_{2\text{max}}$ (l/min)	$\text{VO}_{2\text{max}}$ (ml/kg/min)
Mean	23.4	179.5	75.5	23.4	4.4	57.9
$\pm\text{SE}$	0.8	2.1	3.3	0.6	0.2	1.8

Experimental Procedures

Two counterbalanced trials took place separated by at least a week. On the day of the trial, subjects entered the laboratory in the morning following an overnight fast and at least 16 h after their last exercise bout. They voided and body mass was obtained. A resting blood sample was collected by venipuncture and a probe was inserted 8 cm past the anal sphincter

for measurement of rectal body temperature. The subjects then underwent light exercise on a cycle ergometer (30% $\text{VO}_{2\text{peak}}$) at 40°C and 60% relative humidity until 2.5-3% of initial body mass was lost. Rectal temperature and heart rate were monitored during the dehydration period. Ten of the twelve subjected underwent the same dehydration process described above expect two subjects were dehydrated to similar levels with intermittent exposure to heat and exercise (20 min of light exercise on a cycle ergometer at 20°C followed by 10 min of sauna exposure at 65°C).

After the dehydration period, subjects rested for 30 min to allow the body fluid compartments to stabilize. During this time, subjects changed into dry clothes and had a teflon catheter inserted into an antecubital vein. Following the 30 min transition period, subjects began the 2-hour rehydration period. Subjects were rehydrated with either placebo (CON) or chicken noodle soup (SOUP). The artificially sweetened placebo drink (fruit punch flavored unsweetened Kool Aid) contains no sugar or electrolytes; however, the participants was told they were drinking a sports beverage. Beverage composition and the composition of water are presented in Table 3.

Table 3. Beverage characteristics

Characteristic	CON	SOUP	H ₂ O
Osmolality, mosmol/kg H ₂ O	31.0	338.0	24.5
[Na ⁺], mmol/l	14.4	161.0	3.0
[K ⁺], mmol/l	16.0	5.3	0.0
Total CHO, g/l	-	93.0	-
Simple sugars, g/l	-	3.0	-
Total fat, g/l	-	14.0	-
Total protein, g/l	-	30.0	-

CHO, fat and protein values of SOUP were obtained from data reported in Ray et al. (34). The placebo and soup were administered at 22°C and 50°C, respectively. Subjects ingested 175 ml of the respective beverage at the onset of the rehydration period and 175 ml 20 min later. For the remainder of the rehydration period, all subjects ingested an equal amount of water every 20 min so that the total volume of water ingested was equal to the volume of water lost during dehydration.

After rehydration, subjects performed 30 min of exercise at $67.6 \pm 0.8\%$ $\text{VO}_{2\text{peak}}$ followed by a time trial in which they performed the amount of work that would be performed in 30 min of exercise at 70% $\text{VO}_{2\text{peak}}$ under thermoneutral environment (25°C and 40% relative humidity). Accumulated work was recorded every 5 min during the time trial. Respiratory exchange measurements were taken every 5 min during the exercise bout. Blood samples (8 ml) were drawn immediately after insertion of the catheter, every 20 min during the rehydration period, and every 10 min during the subsequent exercise bout. After each blood draw, the catheter was kept patent with 3 ml of sterile saline solution. Rating of perceived exertion, HR and rectal temperature were monitored throughout post-rehydration exercise. Urine was collected at the end of rehydration, at the end of first 30 min steady state exercise post-rehydration and at the end of the time trial.

Analytical Methods

Hemoglobin (Hb) concentrations were determined using the cyanmethemoglobin method in triplicate. Hematocrit was measured in triplicate using the microhematocrit method. Hematocrit readings were corrected for plasma trapped in red packed red blood cells

(0.96) and also for venous-to-total body hematocrit ratio (0.91) (40). Relative changes in plasma volume were calculated using the method employed by Dill and Costill (15).

Blood was collected in BD Vacutainer® Heparin Tubes. Hemoglobin and hematocrit were measured on the same day of experiment days. The rest of the blood samples were then centrifuged at 2000 rpm for 10 min and plasma was separated. Plasma glucose and lactate were analyzed by Glucose and Lactate analyzer (model 2300, YSI Inc, Yellow Springs, Ohio).

Dietary and Exercise Control

Three-day dietary records were obtained from the subjects prior to the initial trial. The subjects were required to replicate their diet before next trial. In addition, subjects were instructed to drink 1 extra liter of water one day before the trial days to ensure euhydration. Subjects also recorded all physical activities three days before the initial trial and they were required to reproduce the same physical activities before the second trial.

Statistics

Values are presented as means \pm SE. Body mass, oxidation rates of CHO and fat during post-rehydration exercise, blood and urine measurements obtained over time were tested for a treatment by time interaction using a two-way analysis of variance with repeated measures. The accumulated urine volume and total CHO being oxidized during post-rehydration exercise were analyzed by Paired Student t-test. The comparison of trial time during performance test and order effect of time trial was tested by Wilcoxon Rank Sum Test. Differences were considered significant at $P < 0.05$. Effect sizes (Cohen's d) were used

to compare the means. An effect size of 0.2 is indicative of a small effect, 0.5 a medium and 0.8 a large effect size.

CHAPTER 4. RESULTS

Body Mass Changes

The mean body mass loss (Table 4) of the twelve subjects was 2.09 ± 0.27 kg, corresponding to $2.8 \pm 0.2\%$. There were no statistical significant differences in body mass prior to and after dehydration, or percent change in body mass with respect to trial. At the end of the rehydration period, subjects remained somewhat hypohydrated in both CON and SOUP. There was no statistical difference in the percent of body mass loss that was regained at the end of rehydration (CON: $79.4 \pm 3.3\%$; SOUP: $80.9 \pm 3.4\%$).

Table 4. Body mass before and after dehydration, after rehydration and after exercise

Body Mass (kg)	CON	SOUP
Baseline	75.9 ± 2.8	75.5 ± 2.7
Dehydrated	73.8 ± 2.8	73.5 ± 2.7
Rehydrated	75.4 ± 2.8	75.1 ± 2.7
After 30 min steady state exercise	74.8 ± 2.8	74.5 ± 2.7
End	74.0 ± 2.8	73.7 ± 2.7

Mean \pm SE, n=12

Urine volume

Urine volume was lower in SOUP during rehydration, during 30 min steady state exercise and during the time trial (Table 5; significant main effect, $F(1, 11) = 7.21$, $p < 0.05$). Total urine volume was smaller in SOUP compared to CON (266.9 ± 23.3 vs 372.0 ± 46.4 , respectively; $t(11) = 2.69$, $p < 0.05$; effect size=0.6). The effect size of the difference in urine volume measured at the end of post-rehydration exercise was medium ($d=0.5$).

Table 5. Urine volume

Urine Volume	CON	SOUP
During rehydration (ml)	70.0 \pm 12.6	66.8 \pm 11.0
During 30 min steady state exercise (ml)	167.1 \pm 25.7	121.8 \pm 15.5
End (ml)	134.9 \pm 29.1	78.3 \pm 20.5
Total (ml)	372.0 \pm 46.4	266.9 \pm 23.3 *

Urine volumes were measured after rehydration, after 30 min steady state exercise and at the end of time trial. Subjects who were not able to finish time trials, urine were collected at exhaustion. * Significantly different between SOUP and CON ($p < 0.05$). Significantly different between trials (main effect of treatment) Data are means \pm SE, $n=12$

Rehydration

During the rehydration period, the participants drank 434 ± 67 ml of water every 20 min from 40- 120 min of rehydration for a total volume of $1,738 \pm 268$ ml. Plasma glucose concentrations were significantly higher in SOUP during rehydration (Figure 1; significant main effect, $F(1, 11) = 11.51$, $p < 0.05$). There were no statistical significant difference in plasma lactate concentrations between SOUP and CON (Figure 2). When glucose and lactate concentrations were corrected for the changes in plasma volume (39), plasma glucose concentrations were still significantly higher in SOUP ($F(1, 11) = 7.20$, $p < 0.05$). The effect size of the difference in plasma glucose was large at 40 min of rehydration ($d=0.8$), and effect sizes were medium (range: 0.5-0.6) from 20 min of rehydration to 80 min of rehydration. The area under the glucose curve during rehydration was higher in SOUP (456.5 ± 14.8 mmol/L' min vs 488.7 ± 9.3 mmol/L' min, respectively, $t(11) = 2.89$, $p < 0.05$).

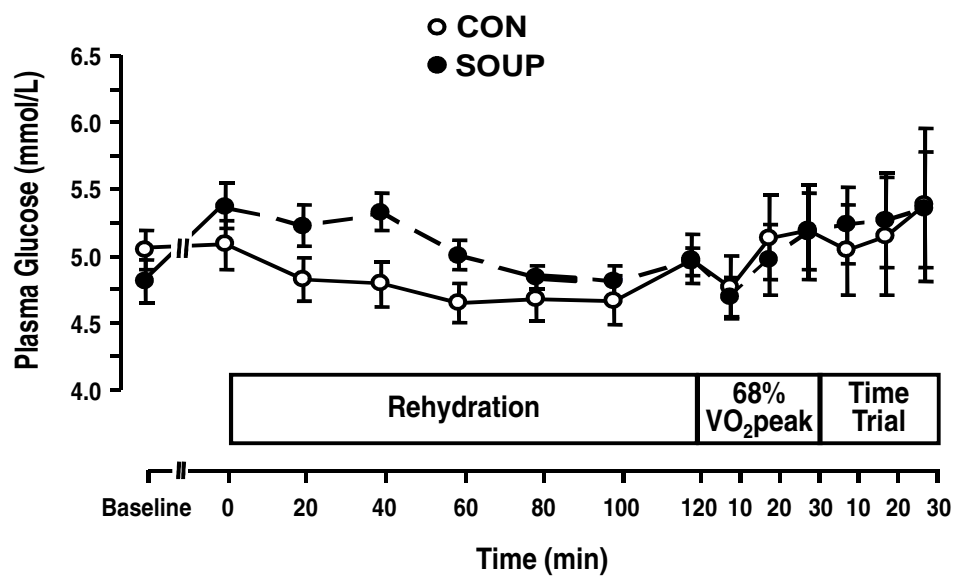


Figure 1. Plasma glucose concentration during rehydration, steady state exercise and time trial up to 30 min for 12 subjects. Significantly greater plasma glucose in SOUP during rehydration (Mean \pm SE, n=12)

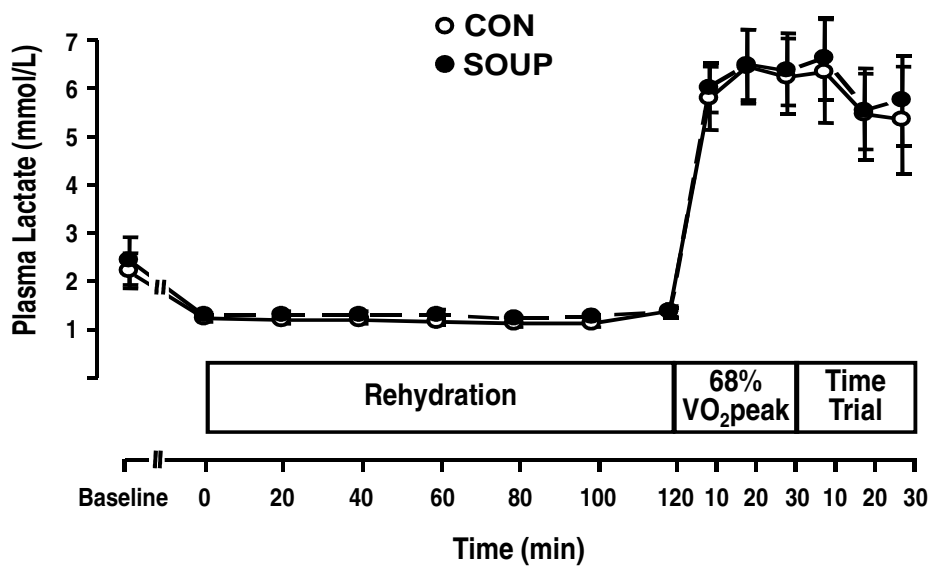


Figure 2. Plasma lactate concentration during rehydration, steady state exercise and time trial up to 30 min (Mean \pm SE; n=12)

Post-rehydration exercise

Plasma volume (Figure 3) decreased with the onset of exercise and the change in plasma volume was not statistically different between CON and SOUP. During steady state exercise and the time trial, plasma glucose and lactate concentrations were not statistically different between CON and SOUP.

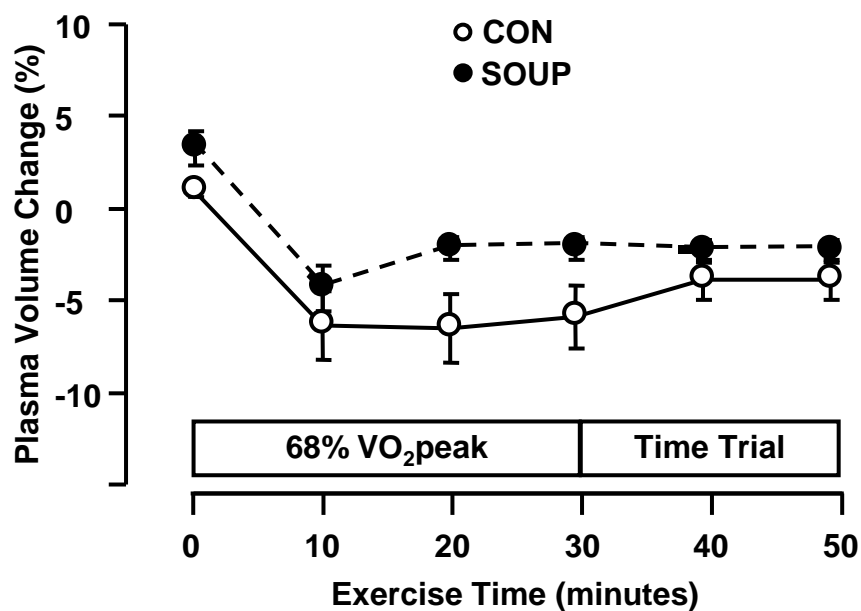


Figure 3. Plasma volume change from the end of rehydration to 20 min in time trial (Mean \pm SE; n=12)

The respiratory Exchange Ratio (RER) was not statistically different between CON and SOUP (Table 6). There was a trend for higher carbohydrate utilization in SOUP during post-rehydration exercise (Figure 4); $F(1, 11) = 3.84$, $p=0.076$. Fat oxidation was not statistically different between CON and SOUP (Figure 5). Total CHO oxidized during post-rehydration exercise was 142.8 ± 19.5 g and 149.5 ± 19.5 g for CON and SOUP, respectively.

Table 6. Respiratory exchange ratios

Exercise time (min)	CON	SOUP
10	0.91 ± 0.01	0.93 ± 0.01
20	0.89 ± 0.01	0.90 ± 0.01
30	0.89 ± 0.01	0.88 ± 0.01
40	0.86 ± 0.02	0.89 ± 0.01
50	0.85 ± 0.02	0.87 ± 0.01
60	0.85 ± 0.01	0.87 ± 0.01

Mean \pm SE; n=12.

Ten subjects finished both steady state exercise and the time trial. One subject was not able to finish the performance test on both trials reaching voluntary exhaustion at 29.5 min in CON and 40.9 min in SOUP. The other subject reached exhaustion at 43.2 min in CON and completed the time trial in 33.4 min in SOUP. Rectal temperatures during post-rehydration exercise were significantly higher in SOUP (Table 7; $p<0.05$). Heart rate and RPE were not statistically different between CON and SOUP. The time to complete the fixed amount of work during the time

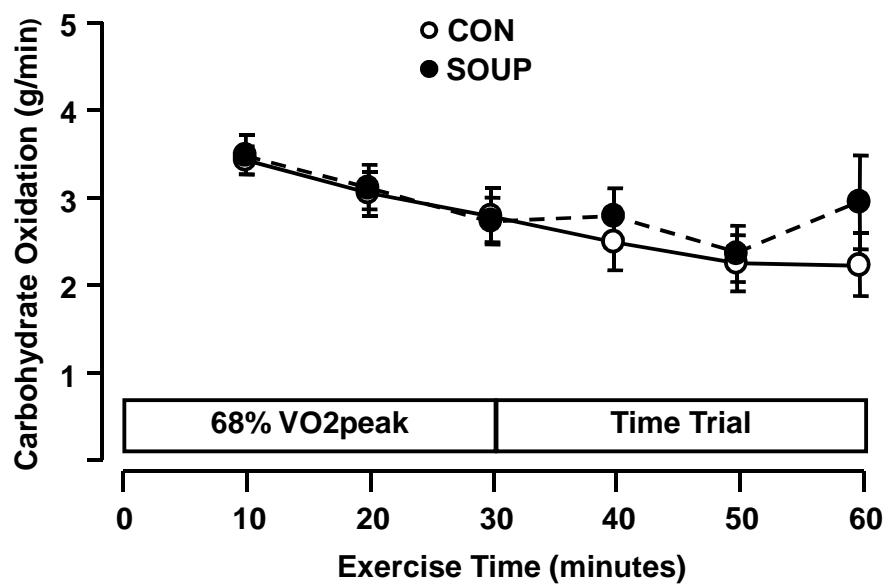


Figure 4. Carbohydrate oxidation rate during post-rehydration exercise (Mean \pm SE; n=12)

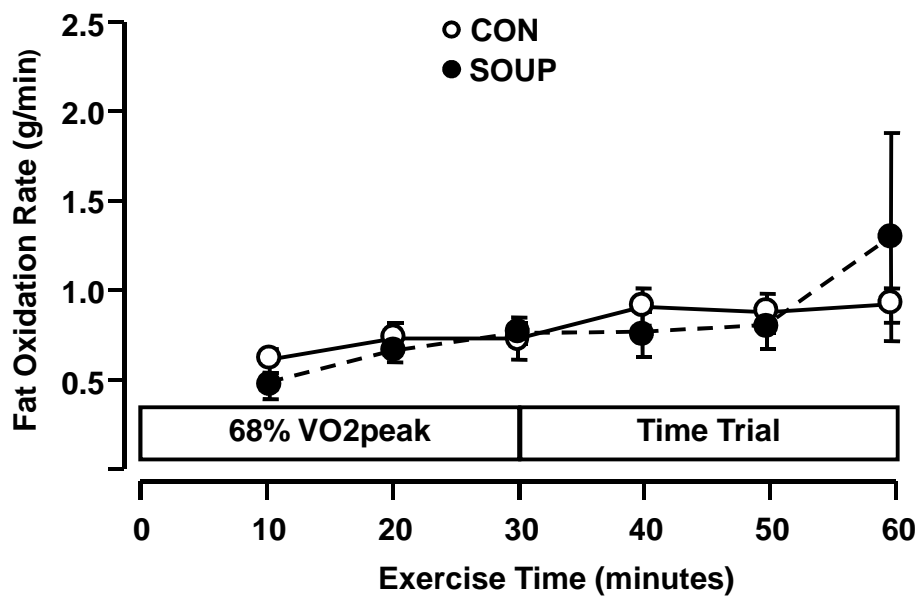


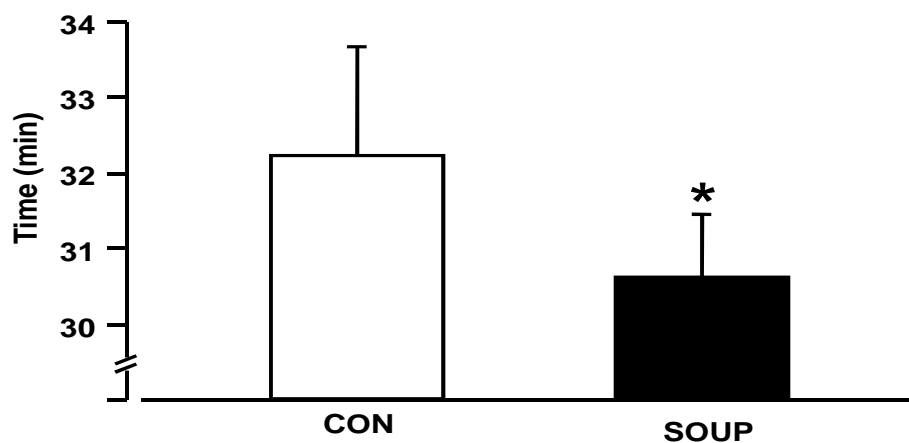
Figure 5. Fat oxidation rate during post-rehydration exercise (Mean \pm SE; n=12)

Table 7. Rectal temperature, heart rate and RPE during post-rehydration exercise

Time (min)	Rectal temperature		Heart rate		RPE	
	CON	SOUP	CON	SOUP	CON	SOUP
10	37.2 \pm 0.1	37.5 \pm 0.1	165 \pm 3	165 \pm 3	14 \pm 1	14 \pm 0
20	37.7 \pm 0.1	37.9 \pm 0.1	169 \pm 4	171 \pm 3	16 \pm 1	16 \pm 0
30	38.1 \pm 0.1	38.3 \pm 0.1	173 \pm 4	174 \pm 4	17 \pm 0	16 \pm 0
40	38.4 \pm 0.1	38.5 \pm 0.1	174 \pm 3	170 \pm 5	18 \pm 0	16 \pm 1
50	38.5 \pm 0.1	38.7 \pm 0.1	173 \pm 5	168 \pm 6	18 \pm 0	17 \pm 0
60	38.6 \pm 0.2	39.0 \pm 0.1	175 \pm 6	167 \pm 8	18 \pm 0	18 \pm 0

Note: Rectal temperatures were significantly different between trials (main effect of treatment). Mean \pm SE; n=12.

trial was significantly shorter (Figure 6; $Z = 1.87$, $p < 0.05$) in SOUP (30.6 ± 0.8 min) than CON (33.2 ± 1.4 min, $n=11$), but the effect size was small (0.3). There was no order effect for performance times ($p=0.477$). Mean power output during the first 25 min was 192.2 ± 13.9 watts in CON and 205.9 ± 10.3 watts during SOUP (Figure 7).



*Significantly different from CON (based on data on 11 subjects, excluding the subject who was not able to finish either CON or SOUP); Mean \pm SE

Figure 6. Trial time during performance exercise (CON: $n=10$; SOUP $n=11$)

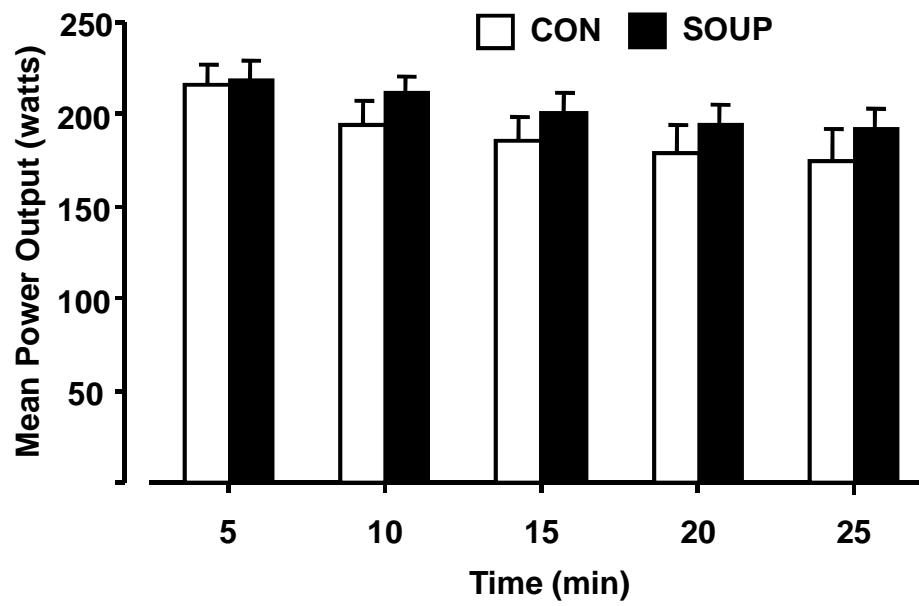


Figure 7. Work output during time trials (only first 25 min shown) (Mean \pm SE; n=12)

CHAPTER 5. SUMMARY AND DISCUSSION

The main finding of this study is that consuming chicken noodle soup following heat and exercise-induced dehydration resulted in a significant improvement in endurance capacity. Complete rehydration was not achieved in either trial. Heart rate, RPE and plasma volume during post-rehydration exercise were not statistically different in SOUP and CON. Finally, rectal temperatures were higher in SOUP during exercise. These findings suggest that the improved performance in SOUP was not likely due to improved hydration status.

This finding of improved endurance capacity after CHO ingestion is in accordance with the findings of Fallowfield et al. (16) and Wong et al. (47) although less CHO was ingested in the present study. In one study (16), subjects ingested either 1 g CHO per kg BM immediately after a 90 min run and 2 h later, while the control group ingested the same amount of water during the 4 h rehydration period. Subjects ran 22 min longer in CHO trial before they reached exhaustion. In another study (47), subjects drank either CHO-electrolyte solution (3.6 g/kg BM) or placebo equal to 200% of the fluid loss during an initial exercise bout. During a second exercise bout to exhaustion, subjects ran 24 min longer when fed CHO. Studies suggest that CHO intake as small as 50 g CHO during rehydration has similar effect as 170 g on subsequent exercise run time to exhaustion (0.7 and 2.1 g/kg BM respectively)(11).

The improved post-rehydration exercise performance in the present study may be attributed to greater carbohydrate availability. The CHO oxidation rate during post-rehydration exercise, although not statistically different, tended to be higher in the SOUP compared to CON. Estimated total CHO oxidized based on indirect calorimetry was not

statistically different between CON and SOUP. It should be noted that during performance exercise, rather than exercising at the same intensity, subjects were exercising at the intensity they felt comfortable and it might vary from minute to minute. At the time when respiratory data were collected the subjects may be exercising at an intensity higher or lower than the average workrate during the 10 min interval. Therefore, CHO oxidation rates might be overestimated or underestimated.

According to Bergström and Hultman, muscle glycogen is depleted at a rate of approximately 19 mmol/ kg muscle during 100 min exercise at 30% $\text{VO}_{2\text{peak}}$. Exercise performed in hot environment increases muscle glycogen utilization by 76% (20). Therefore, muscle glycogen used during dehydration may be 33 mmol/ kg muscle, corresponding to a 1/3 decline in original muscle glycogen store, assuming baseline muscle glycogen store is approximately 100 mmol/kg muscle.

Although liver or muscle glycogen was not measured in the present study, there were a few studies on liver and muscle glycogen restorations rates after feeding. Previous studies have demonstrated that short-term skeletal muscle glycogen resynthesis after exercise-induced glycogen depletion is linearly related to the exogenous CHO load, when the CHO load is relatively high (12 g/kg BM) (43). Blom et al. (5) found muscle glycogen resynthesis rates of $2.1 \text{ mmol} \cdot \text{kg wet wt}^{-1} \cdot \text{h}^{-1}$, during feedings of 0.35g/kg BM glucose given at 2-h intervals for 6-h recovery period after exercise-induced glycogen depletion. During rehydration period, the amount of CHO contained in SOUP in the present study was 33.6 g, approximately 0.4 g/kg BM. Therefore, the muscle glycogen resynthesis rate would be slightly higher than $2 \text{ mmol} \cdot \text{kg wet wt}^{-1} \cdot \text{h}^{-1}$ in the present study. Calculated glycogen stored in the active muscle averaged approximately $4 \text{ mmol} \cdot \text{kg wet wt}^{-1}$ during 2 h rehydration

period. The amount of muscle glycogen utilized during the dehydration exercise was almost seven-fold compared to the estimated amount of muscle glycogen resynthesized during recovery period. It was not likely to fully restore muscle glycogen within 2 h with 33.6 g CHO.

Casey et al. (6) did not find significant changes in muscle glycogen content after CHO ingestion. It was speculated that this finding was due to relatively small CHO load (1 g/ kg BM) and a portion of the CHO was extracted by the liver (30%) before being made available to the large muscle mass. When the change in muscle glycogen and liver glycogen were considered together, the sum was significantly correlated with CHO intake. During the rehydration period, the amount of CHO contained in SOUP in the present study was lower than the amount administered in Casey et al. (6). Therefore, it is likely that muscle glycogen was not improved in SOUP compared to CON.

Prolonged starvation or a carbohydrate-poor diet decreases liver glycogen from 232 to 24-55 mmol glucosyl units per kg within 24 h. Refeeding with a carbohydrate-rich diet increases liver glycogen to supernormal values (426-624 mmol glucosyl units per kg wet liver tissue) (31). The rate of liver glycogen resynthesis during the first 24 h after refeeding with high CHO diet was $16.7\text{--}23.7 \text{ mmol}\cdot\text{kg wet wt}^{-1}\cdot\text{h}^{-1}$. There is also evidence that prior exercise may facilitate splanchnic glucose output after glucose ingestion (75 g), resulting in a greater rate of glucose appearance and glucose uptake (37). It was suggested by the authors that the heightened rate of appearance of oral glucose was due to adaptations in splanchnic tissues by prior exercise because there was no difference in plasma insulin or glucose clearance rate between CHO trials and control trials. A possible adaptation might be a facilitated delivery of ingested glucose to the systemic circulation.

The blood glucose concentration after administration of oral CHO is dependant on the release of glucose to the intestine, the transport over the intestinal mucosa to the blood and glucose uptake into the muscle cells. The higher plasma glucose concentrations during rehydration in SOUP in which CHO was ingested was in agreement with previous studies (4, 38, 47). The CHO dose used in previous studies ranged from 50 g to 220 g, which was greater compared to the present study (33.6 g; 0.4 g/kg BM). It is surprising that relatively small amount of CHO used in the present study still had an effect on recovery plasma glucose concentration. Several studies found no significant difference between CHO ingestion trials and control trials (16, 42). In these studies (16, 47), elevated insulin was observed after CHO ingestion.

Coyle et al. (14) examined the effect of a high CHO meal (2g/ kg BM) on substrate usage during prolonged exercise 4 h after feeding. Although insulin returned to basal levels before beginning exercise, there was still a rapid decline in plasma glucose concentration and a suppression of lipolysis during the 1st h of exercise suggested being due to persistent effect of insulin. In the present study, CHO intake was lower, but the time between CHO intake and exercise was only 2 h. There were slight decline during the onset of exercise in plasma glucose in both SOUP and CON, suggesting ingesting small amount of CHO had little, if there was any, effect on insulin 2 h after feeding.

A another study by Coyle et al. (13) suggested when muscle glycogen levels were low after 3 h strenuous exercise, subjects who ingested CHO (2.0g/ kg BM at 20 min and 0.4g/ kg BM every 20 min thereafter) were able to maintain plasma glucose at 4-5 mmol/L and exercise for an additional hour compared to controls. The higher CHO oxidation

observed in CHO feeding trials was mainly derived from blood glucose during the 3 to 4 h exercise.

In the present study, it can be speculated that the exogenous CHO from chicken noodle soup may have replenished, at least partly, muscle and liver glycogen during rehydration, even though muscle glycogen level would still be low based on the calculations. During post-rehydration exercise, splanchnic tissues may facilitate higher splanchnic glucose output and higher blood borne substrates might be utilized during SOUP.

Several studies have suggested that adding PRO to CHO supplementation after exercise may help to promote greater recovery of muscle glycogen and attenuate muscle damage. Ivy et al. (25) instructed cyclists to complete a 2.5 h bout of intense cycling before ingesting either a CHO + PRO + Fat (80 g CHO, 28 g PRO, 6 g Fat), low CHO (80 g CHO, 6 g fat), or a high CHO (108 g CHO, 6 g fat) supplement immediately after exercise, and 2 h post-exercise, to determine if the CHO + PRO + Fat combination promoted greater restoration of muscle glycogen. While glycogen replenishment did not differ between the two CHO conditions (low CHO [70.0 ± 4.0 mmol/kg/wet wt] and high CHO [75.5 ± 2.8 mmol/kg/wet wt]), muscle glycogen levels were significantly greater ($p < 0.05$) in the CHO + PRO + Fat treatment (88.8 ± 4.4 mmol/kg/wet wt). The authors suggested that a CHO + PRO + Fat supplement may be more effective because of its provocation of a more pronounced insulin response. Similarly, Berardi et al. (1) utilized cyclists for the completion of exercise bouts of 60 – 90 min on separate occasions before ingesting CHO + PRO or CHO alone. CHO+PRO supplements, given early after exercise, enhance glycogen resynthesis relative to CHO and placebo. However, performance was not influenced in this type of

exercise bout. Given the low protein dose in the current study (10.5 g), protein may not likely to enhance glycogen resynthesis rate.

The addition of protein during recovery may due to the additional energy it provides. Betts et al. (3) recruited six active males for three trials, each involving a 90-min treadmill run at 70% maximal oxygen uptake (run 1) followed by a 4-h recovery. At 30-min intervals during recovery, participants ingested solutions containing: (1) 0.8 g carbohydrate per kg body mass (BM) per h plus 0.3 g kg BM⁻¹ h⁻¹ of whey protein isolate (CHO-PRO); (2) 0.8 g carbohydrate kg BM⁻¹ h⁻¹ (CHO); or (3) 1.1 g carbohydrate kg BM⁻¹ h⁻¹ (CHO-CHO). Following recovery, participants ran to exhaustion at 70% maximal oxygen uptake (run 2). Exercise capacity during run 2 was greater following ingestion of CHO-PRO and CHO-CHO than following ingestion of CHO with no significant difference between the CHO-PRO and CHO-CHO treatments. Increasing the energy content of these recovery solutions extended run time to exhaustion, irrespective of whether the additional energy originated from sucrose or whey protein isolate. Therefore, the amount of protein in the chicken noodle soup, although small, added energy content in this beverage. Further research is need to determine this beneficial effect.

In a previous study in our laboratory using similar rehydration protocols (34), higher plasma volume restoration and lower urine volumes were observed with rehydration food or beverages containing higher sodium concentrations (109.5 mmol/l). An earlier study by Nose et al. (33) showed faster plasma volume restoration and lower urine output in subjects consuming water and capsules containing sodium (77 mmol/L). In contrast, urine volumes were not statistically different between CHO-electrolytes drinks and water intakes after exercise-induced dehydration (47). The differences in urine output might be explained by the

lower amount of sodium in the CHO-electrolyte beverage (24 mmol/L). The large amount of fluid (88.5-200% BM loss) ingested during rehydration might also mask the positive effect of sodium in retaining body fluid. Although urine outputs were not statistically different between two trials, there was a trend of lower volumes in CHO-electrolytes solution groups. In the present study, significant lower urine output was observed in SOUP than in CON. This finding was in agreement with previous studies (33, 34, 47). The results suggest that greater fluid retention was achieved in SOUP. However, greater fluid retention did not result in larger body mass in SOUP. This may be because the difference in urine volume (0.12 kg) was too small to cause any significant difference in body mass.

In conclusion, although only 350 ml of treatment beverage was ingested at the beginning of rehydration, chicken noodle soup improved in restoring endurance capacity after 2 h rehydration period. There was a trend of higher CHO oxidation rate in post-rehydration exercise; therefore, better performance in SOUP might due to greater CHO availability. Muscle glycogen may not be fully restored at the end of rehydration, greater CHO oxidation might be due to greater blood-borne substrates and hepatic glucose output in SOUP.

APPENDIX A. HUMAN SUBJECTS AND INFORMED CONSENT

IOWA STATE UNIVERSITY OF SCIENCE AND TECHNOLOGY

DATE: 15 January, 2008

TO: Dr Douglas S. King
248 Forker Bldg

Rick L. Sharp
250 Forker Bldg.

FROM: Jan Canny, IRB Administrator
Office of Research Assurances

TITLE: **The Effect of Sodium and Carbohydrate in a Rehydration Beverage when Consumed as a Mean on Subsequent Exercise Performance**

Institutional Review Board
Office of Research Assurances
Vice President for Research
1138 Pearson Hall
Ames, Iowa 50011-2207

515 294-4566
FAX 515 294-4267

IRB ID: 07-605

Approval Date: 9 January 2008

Date for Continuing Review: 4 December 2008

The Institutional Review Board has reviewed and approved this project. Please refer to the IRB ID number shown above in all correspondence regarding this study.

Your study has been approved according to the dates shown above. To ensure compliance with federal regulations (45 CFR 46 & 21 CFR 56), please be sure to:

- **Use the documents with the IRB approval stamp** in your research
- **Obtain IRB approval prior to implementing any changes** to the study by completing the "Continuing Review and/or Modification" form
- **Immediately inform the IRB of (1) all serious and/or unexpected adverse experiences** involving risks to subjects or others; and (2) **any other unanticipated problems involving risks** to subjects or others.
- **Stop all research activity if IRB approval lapses**, unless continuation is necessary to prevent harm to research participants. Research activity can resume once IRB approval is reestablished.
- **Complete a new continuing review form** at least three to four weeks prior to the **date for continuing review** as noted above to provide sufficient time for the IRB to review and approve continuation of the study. We will send a courtesy reminder as this date approaches.

Research investigators are expected to comply with the principles of the Belmont Report, and state and federal regulations regarding the involvement of humans in research. These documents are located on the Office of Research Assurances website [www.compliance.iastate.edu] or available by calling (515) 294-4566.

Upon completion of the project, please submit a Project Closure Form to the Office of Research Assurances, 1138 Pearson Hall, to officially close the project.

INFORMED CONSENT

Title of Study: The effect of sodium and carbohydrate in a rehydration beverage when consumed as a meal on subsequent exercise performance.

INVESTIGATORS: DOUGLAS S. KING, PHD RICK L. SHARP, PHD

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This is a research study. Please take your time in deciding if you would like to participate. Please feel free to ask questions at any time.

INTRODUCTION

Research has investigated factors that optimize fluid absorption and retention leading to restoration of body weight and plasma volume after heat and/or exercise induced dehydration. Ingestion of fluids (soup) containing both carbohydrates and high concentrations of electrolytes (especially sodium) appear to restore plasma volume while reducing the amount of fluid lost in the urine when compared to ingesting only water or fluids containing low concentrations of sodium. Whether consumption of a soup will increase an individual's ability to perform exercise after a period of rehydration is unknown. Also, subsequent exercise performance improvement may be the result of increased muscle glycogen concentrations because of carbohydrate ingestion and not due to improved hydration status. This project is designed to determine 1) whether this soup optimizes rehydration compared to a sports drink after exercise induced dehydration, 2) whether rehydration with this soup results in improved cardiovascular function, temperature regulation, and metabolism, and 3) whether any improvement in exercise performance occurs after improved hydration status.

DESCRIPTION OF PROCEDURES

Before participating in this research study please carefully read the following procedures and ask questions if needed. *If you decide you would like to participate in this study after carefully reading this document, you will be asked to complete a medical history questionnaire.* The medical history form contains general information on your past medical history and current health status including physical activity (type and amount). Only after the informed consent is signed and the medical history questionnaire is reviewed and approved will you be allowed to participate in this study.

Previous health conditions that will warrant your exclusion include: overall poor health, history of high blood pressure, and/or heart attack. Only college-aged (18 - 35 years old), non-smoking, non-diabetic men not on any type of medications that affect water regulatory hormones will be allowed to participate in this study. You will also be excluded if you are not physically active (less than 3-4 days of physical activity per week for less than 30 minutes). An allergy to Nutrasweet (aspartame) is also grounds for exclusion. Women will not be recruited for this study because of potential confounding factors associated with their menstrual cycles.

Before the first trial, you will participate in a graded exercise test for determination of your VO₂peak. **In the graded exercise test you will be asked to ride a stationary cycle at progressively harder workloads until the maximal amount of oxygen your body can utilize is reached (VO₂peak).** This measurement will be used for the assignment of workrates during the dehydration period and subsequent performance trial.

Two randomly assigned trials will take place separated by at least a week. **Prior to each trial, you will be asked to record a three day diet record (food and fluid) and exercise diary that will be replicated prior to the other trial. To ensure proper hydration on testing days, we ask that you drink at least one extra liter of water the day before trials. Each trial is expected to last approximately 5 hours.**

On the day of the trial, you will enter the laboratory at 7:00 am following an overnight fast and at least 16 hours after your last exercise bout. You will void and body weight will be obtained. A resting blood sample will be collected by venipuncture, and you will insert a probe 8 cm past the anal sphincter for measurement of rectal body temperature. The venipuncture will be performed by trained personnel (Zeb Sullivan, Dr. Rick Sharp or Dr. Douglas King).

You will then undergo an exercise session **on a stationary cycle** at 30% VO₂peak in a climatic chamber set at 40°C (104°F), 60% Relative Humidity (RH) until 2.5 - 3.0% of initial body weight is lost (60-90 minutes anticipated). Rectal temperature, heart rate (HR), ratings of perceived exertion (RPE), and respiratory exchange measurements will be taken every 10 minutes.

Immediately after the dehydration period, **a muscle biopsy will be performed on the lateral aspect (outside) of your vastus lateralis muscle (thigh) under local anesthetic. After your leg has been cleaned with surgical scrub, a small amount of xylocaine (a local anesthetic) will be injected into the skin and tissue surrounding the muscle. After allowing several minutes for the anesthetic to take effect, a small incision (½ inch long) will be made through your skin and connective tissue. The muscle sample will then be removed by inserting a muscle biopsy needle through the incision into the muscle and snipping off a piece of muscle that is approximately the size of a pea.**

During the biopsy procedure, the most common sensation is a slight burning that occurs when the anesthetic is given, and which disappears within a few seconds. Many subjects also experience a “tugging” sensation when the small piece of muscle is removed.

You will undergo a total of six muscle biopsies (three each trial); one immediately following exercise induced dehydration, one after rehydration, and one following 30 minutes of exercise at 70% of VO₂peak. The muscle biopsies will be used to assess changes in total muscle water and glycogen concentration. All biopsies will be performed by Rick L. Sharp, Ph.D. or Douglas S. King, Ph.D. under sterile conditions.

A pressure bandage will be placed over the incision site **following the first muscle biopsy** and you will be allowed to rest for 30 minutes to allow the body fluid compartments to stabilize.

During this time you may shower and change into dry clothes. During this 30 minute transition period you will also have a small flexible catheter inserted into your antecubital vein for later blood sampling. Following the 30-minute transition, you will begin the 2-hour rehydration period in which either a sports beverage (SB) or chicken noodle soup (CNS) will be consumed. The SB and soup will be administered at 22°C (72°F) and 50°C (120°F) respectively. One-hundred seventy-five milliliters (ml) of the respective beverage will be consumed at the beginning of the rehydration period, and 175 ml twenty minutes later. For the remainder of the rehydration period, you will ingest an equal amount of water every 20 minutes so that the total volume of water ingested is equal to the volume of water lost during dehydration.

Immediately after the rehydration period, a small incision (0.5 inch) will be made on your left vastus lateralis and a muscle biopsy will be taken. A pressure bandage will be placed over the incision site and you will perform 30 minutes of exercise at 70% VO₂peak. The final muscle biopsy will be taken from the incision on the left vastus lateralis following the 30 minutes of exercise at 70% VO₂peak. After the last biopsy, you will perform a time trial in which you will be asked to complete the same amount of work (70% VO₂peak, 30 minutes) as fast as possible. The second exercise bout and time trial will each be conducted in the climatic chamber set at room temperature (77°F) and humidity (40%). Rectal temperature, heart rate, ratings of perceived exertion, and respiratory exchange measurements will be recorded every 10 minutes during the dehydration bout (first exercise session) and every 5 minutes during the second exercise bout and time trial.

Blood samples (8 ml) will be drawn immediately after insertion of the catheter (during the transition period), every 20 minutes during the rehydration period, and every 10 minutes during the subsequent exercise bout and time trial (total of 15 blood samples or about 4 ounces) and analyzed for hemoglobin, hematocrit, osmolality, and plasma sodium, potassium, glucose, and lactate concentrations. Catheter placement and blood sampling will be performed by trained personnel (Zeb Sullivan, Dr. Douglas King, or Dr. Rick Sharp). After each blood draw, the catheter will be kept patent with 1-3 ml of sterile saline solution. Muscle biopsies will be taken after the dehydration period, after the rehydration period, and after 30 min of exercise (total of 3 muscle biopsies). Urine will be collected upon entering the lab, after dehydration, after rehydration, and after the time trial (4 collections). All urine collected during the trial will be analyzed for total urine volume, specific gravity, osmolality, and sodium and potassium concentrations.

RISKS

The muscle biopsies are associated with some risk to the subjects, including infection of the biopsy site and some degree of mild soreness the day after the biopsy. These risks are minimized by using sterile procedures and instruments, placing slight pressure over the biopsy site for 5 minutes after the biopsy, and by applying a pressure bandage over the site for 12 hours following the biopsy. *In the more than 3,000 muscle biopsies performed by Drs. King and Sharp, who will perform biopsies in this study*, there has been only one instance of infection. Delayed and minor soreness has been reported in ~10% of the subjects. In no instances have subjects reported an amount of soreness sufficient to affect their usual daily activities.

A very rare side effect of the muscle biopsy is lightheadedness or dizziness. This is seen in less than 1% of subjects, and subsides within a few minutes of the procedure. Elevation of the legs rapidly stops this side effect.

Blood sampling and infusion into forearm veins using a 1 inch polyethylene catheter is associated with a risk of infection, hematoma, and transmission of communicable diseases. These risks are minimized by using sterile procedures, applying adequate pressure over the puncture site following withdrawal of the catheter, and the use of disposable equipment including catheters and

sterile latex gloves. All catheters will be inserted by trained personnel (Zeb Sullivan, Dr. Rick Sharp or Dr. Douglas King). The risk associated with infusing saline into human subjects is considered to be minimal. Sterile procedures will be used in the handling of all infusates to minimize the risk of infection.

The dehydration and subsequent exercise performance regimens may result in slight muscle soreness, localized to the muscles used in the exercise program. This soreness should in no way result in the inability of the subjects to engage in their normal daily activities. The exercise program may produce light headedness, dizziness, and in extremely rare instances, myocardial infarction. These risks will be minimized by selecting young healthy subjects (you) with little risk for a cardiovascular incident, and by the careful supervision of all exercise sessions by knowledgeable and CPR-trained individuals. Two automated external defibrillators (AEDs) are located near the exercise metabolism laboratory. The risk of heat illness is small, since the level of dehydration is mild, and you are in good physical condition. In addition, exercise and/or heat exposure will be determined if the heart rate exceeds 180 beats/min or if the rectal temperature exceeds 39.5 C.

The risk during the administration of the supplemental rehydration beverages include contamination of the beverages and/or glassware with dirt, bacteria, or blood is minimal. In order to prevent this possibility, we will properly sterilize all glassware and separate the beverages from contaminated materials. In the event that there is evidence of adverse side effects, you will be immediately informed and the trial will be halted. You will then be immediately referred to the student health center for follow up treatment to ensure that the situation is reversed.

BENEFITS

The information gained by this research will benefit individuals who are at risk for dehydration and heat stress, either by virtue of their participation in sports, or as part of their employment. After completion of this study, we will be able to make recommendations as to whether a soup containing carbohydrate and sodium improves hydration status compared to a sports beverage and, if hydration is improved, whether it increases the ability to perform a subsequent exercise bout. Participants involved in this study will benefit by learning their maximal oxygen consumption, body composition, how well they rehydrate, and whether a sports beverage or soup enables them to perform better after soup consumption.

COSTS AND COMPENSATION

You will be compensated for participating in this study. Volunteers will be paid \$125 for each trial for a total of \$250 for the completion of the study. If you withdraw from the study, you will receive \$125 for the trial completed.

You will need to provide your social security number (SSN) and address in order for us to pay you. This information allows the University to fulfill government reporting requirements and confidentiality measures are in place to keep this information secure. You are given the opportunity to forego receipt of payment(s) and continue in the research study if you decline to provide your social security number and address.

PARTICIPANT RIGHTS

Your participation in this study is completely voluntary and you may refuse to participate or leave the study at any time. If you decide to not participate in the study or leave the study early, it will not result in any penalty or loss of benefits to which you are otherwise entitled.

At any time during the study, you may withdraw your consent to participate without prejudice toward you. Such withdrawal can be for any reason you choose. Constant monitoring of all experiments will be performed by knowledgeable and CPR trained individuals in an attempt to prevent any complications. Emergency first aid supplies and equipment will be immediately available.

RESEARCH INJURY

Emergency treatment of any injuries that may occur as a direct result of participation in this research is available at the Iowa State University Thomas B. Thielen Student Health Center, and/or referred to Mary Greeley Medical Center or another physician or medical facility at the location of the research activity. Compensation for any injuries will be paid if it is determined under the Iowa Tort Claims Act, Chapter 669 Iowa Code. Claims for compensation should be submitted on approved forms to the State Appeals Board and are available from the Iowa State University Office of Risk Management and Insurance.

CONFIDENTIALITY

Records identifying participants will be kept confidential to the extent permitted by applicable laws and regulations and will not be made publicly available. However, federal government regulatory agencies auditing departments of Iowa State University and the Institutional Review Board (a committee that reviews and approves human subject research studies) may inspect and/or copy your records for quality assurance and data analysis. These records may contain private information.

All data and samples will be coded numerically by subject and no names, initials, or other identifying characteristics will be reported in publication or presentation.

QUESTIONS OR PROBLEMS

You are encouraged to ask questions at any time during this study.

- For further information about the study please contact

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- If you have any questions about the rights of research subjects or research-related injury, please contact the IRB Administrator, (515) 294-4566, IRB@iastate.edu, or Director, Office of Research Assurances, (515) 294-3115, 1138 Pearson Hall, Ames, Iowa 50011.

PARTICIPANT SIGNATURE FOR STUDY

Your signature indicates that you voluntarily agree to participate in this study, that the study has been explained to you, that you have been given the time to read the document and that your questions have been satisfactorily answered. You will receive a copy of the written informed consent prior to your participation in the study.

Participant's Name (printed) _____

(Participant's Signature)

(Date)

PARTICIPANT SIGNATURE FOR MUSCLE BIOPSIES

Your signature indicates that the muscle biopsy procedures within this study have been carefully explained to you, that you have read and understand the muscle biopsy procedures, any questions you have about the biopsy procedures have been satisfactorily answered, and you are giving consent for muscle biopsies to be performed in this study.

Participant's Name (printed) _____

(Participant's Signature)

(Date)

INVESTIGATOR STATEMENT

I certify that the participant has been given adequate time to read and learn about the study and all of their questions have been answered. It is my opinion that the participant understands the purpose, risks, benefits and the procedures that will be followed in this study and has voluntarily agreed to participate.

(Signature of Person Obtaining Informed Consent)

(Date)

APPENDIX B. MEDICAL HISTORY QUESTIONNAIRE

Please respond to the following items as accurately as possible. This information will be used by the investigator to ensure a safe exercise environment and to determine if there are any contraindications to exercise or participation in this study. All information will remain confidential unless further professional consultation is warranted.

A) Personal Information

Name _____ Age _____ Tel. # _____
 Height _____ Weight _____ Male or Female (circle)

B) Medical Information

- How would you describe your recent general health?
 _____ Excellent _____ Good _____ Fair _____ Poor
- Place an X in those boxes which describe symptoms or disorders which you have been diagnosed to have. If possible, also indicate the date of the diagnosis.

_____ high blood pressure	_____ arthritis	_____ chest pain
_____ irregular heart beat	_____ epilepsy	_____ heart attack
_____ heart murmur	_____ anemia	_____ migraine
_____ asthma	_____ back trouble	_____ headaches
_____ hay fever or other allergies	_____ dizziness/	_____ diabetes
_____ other _____	_____ fainting spells	
- Describe any surgery that you have had within the last two years: _____.
- Have you ever sustained an injury or experienced any type of chronic pain which has been diagnosed as due to physical activity or sports participation?
 _____ Yes _____ No
 If yes, please describe _____
 How long ago? _____
- Do you smoke cigarettes? _____ Yes _____ No
- Are you presently taking any of the following medications?

_____ drugs to control blood pressure	_____ drugs for asthma
_____ drugs to regulate heart rate	_____ drugs for diabetes
_____ drugs for allergies	_____ thyroid hormone
_____ cortisone	_____ prednisone

Indicate the name(s) of those drugs _____
 Also note the dosage and frequency of use _____
- How long has it been since your last physical examination?
 _____ less than 1 year _____ 1-2 years _____ 2-3 years
 _____ more than 3 years
- Have any of the above symptoms, disorders or injuries limited your physical activity in the past?

 In what way? _____

9. Do you have any food or drug allergies? If yes, please indicate the food or drug below
_____.

10. Are you allergic to Nutrasweet (aspartame) or any other artificial sweetener?
_____.

C) Family Medical History

1. Have any of your blood relatives been diagnosed as having any of the following symptoms/disorders? (Include grandparent, parents, brothers, sisters)

_____ heart attack, under age 50	_____ asthma or hay fever
_____ stroke, under age 50	_____ congenital heart disease
_____ high blood pressure	_____ heart surgery
_____ hyperlipidemia (high cholesterol)	_____ diabetes
_____ obesity	
_____ other _____	

D) Exercise Information

1. List and give the date of any supervised exercise or sports program that you have participated in recently _____

2. Are you currently participating in a regular program of physical activity?

_____ Yes _____ No.

If yes, how often do you exercise per week (on average)?

_____ 1-2 days/wk	_____ 5-6 days/wk
_____ 3-4 days/wk	_____ every day

For how long do you exercise each day?

_____ < 30 min/day	_____ 30-60 min/day
_____ 60-90 min/day	_____ 90-120 min/day
_____ > 120 min/day	

What types of activities are regularly included in your program?

_____ jogging	_____ calisthenics
_____ cycling	_____ swimming
_____ weight lifting	_____ aerobic dance
_____ recreational sports (basketball, racquetball, volleyball, tennis, etc)	
_____ other _____	

How long have you been in your present program?

_____ less than 1 month	_____ 6 months to 1 year
_____ 1-3 months	_____ more than 1 year
_____ 3-6 months	

3. How would you categorize your current physical fitness level?
_____ superior _____ good _____ average _____ below average _____ poor
4. Is there any reason why you think your activity should be limited in this research project?

I attest that all of the above information is accurate to the best of my knowledge

Signed _____ Date _____

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ACKNOWLEDGMENTS

I would like to take this opportunity to express my thanks to those who helped me with various aspects of conducting research and the writing of this thesis. At the very first, I'm honored to express my deepest gratitude to my dedicated supervisor, Dr. Douglas S. King. He has offered me valuable ideas, suggestions and criticisms with his profound knowledge and rich research experience. I'm also extremely grateful to my committee members, Drs. Rick L. Sharp and Katherine T. Thomas, whose patient and meticulous guidance and invaluable suggestions were indispensable to the completion of this thesis. This thesis research was conducted with the assistance and support of many people in the Department of Kinesiology. Special thanks to Dr. Jerry Thomas for statistical advice, Mr. Todd Weber, and Mr. Zebblin Sullivan for conducting this research together.

I express my sincere gratitude towards my family, who have born a silent pain to be apart, but still encouraged me to excel in my field.